Pseudechiniscus asper, a new Tardigrada (Heterotardigrada: Echiniscidae) from Hokkaido, northern Japan

Wataru Abe, Kazuo Utsugi, and Masatsune Takeda

 (WA) Department of Biological Sciences, Graduate School of Science, The University of Tokyo.
(Corresponding address: Department of Zoology, National Science Museum, 3-23-1 Hyakunincho, Shinjuku-ku, Tokyo, 169-0073 Japan);

 (KU) Hiraoka Environmental Science Laboratory, 8-8 Hosoyama, Asao-ku, Kawasaki-shi, Kanagawa, 215-0001 Japan; (MT) Department of Zoology, National Science Museum, 3-23-1 Hyakunincho, Shinjuku-ku, Tokyo, 169-0073 Japan

Abstract.—A new semi-terrestrial tardigrade species of the family Echiniscidae, *Pseudechiniscus asper*, is described from a single male collected from Hokkaido, northern Japan. This species can be readily distinguished from the known congeners by a combination of the following characters: almost the entire surface of all dorsal plates is covered with coarse cuticular granulation; the subtriangular projection is present at position D; paired lobes on pseudosegmental plate are roundly triangular and tipped with papillate projection; and, each internal claw is armed with a spur.

In autumn of 1988, the second author collected lichen samples to obtain the tardigrades around Lake Shikotsu in southwestern Hokkaido, northern Japan. Tardigrade specimens extracted from the lichen samples were fixed with 5% formalin, and mounted on glass slides with Hoyer's medium. Cover slips were sealed by Paraffin-Balsam. Close examination was performed using a phase contrast and Nomarski differential interference microscope (ZEISS Axiophoto) and illustrations were depicted with the aid of camera lucida.

After a review of the literature, it was concluded that one of the specimens represents a new species belonging to the genus *Pseudechiniscus* of the family Echiniscidae, which is described and illustrated as *Pseudechiniscus asper*, new species. Terminology is mainly that used in Dastych & Kristensen (1995) and Kristensen (1987).

Pseudechiniscus asper, new species Figs. 1-2

Type material.—Holotype: Four-clawed adult male; Shikotsuko-onsen, Chitose,

southwestern Hokkaido, northern Japan (141°24'N, 42°46'E); ca. 250 m; extracted from a foliose lichen, *Phaeophyscia imbricata*, on trunk of *Acer japonicum*, 9 Oct 1988, K. Utsugi leg. Holotype is deposited in the National Science Museum, Tokyo (NSMT), under the registered number NSMT-Tg 44.

Diagnosis.—Small size. Dorsal plates covered with coarse cuticular granulation. Scapular plate and segmental paired plates II and III each with segmental lateral plates. Subtriangular projection at position D. Paired lobes on pseudosegmental plate roundly triangular, tipped with papillate projection. Internal claws each with a spur.

Description.—Body length 166 μ m excluding leg IV, width 83 μ m; body width: body length, 1:2. Eyespot not detected, body color evenly translucent in Hoyer's medium.

Dorsal plates well developed and conspicuous. Delineation of dorsal plates and subdivision within a plate each comprised of cuticular ridge. Almost entire surface of

PROCEEDINGS OF THE BIOLOGICAL SOCIETY OF WASHINGTON



Fig. 1. *Pseudechiniscus asper*, new species, holotype male (NSMT-Tg 44), Hokkaido, N Japan: A, Habitus, dorsal view; B, Habitus, ventral view; C, Internal claw of leg II; D, External claw of leg II. Abbreviations: cr = cuticular ridge; D = subtriangular projection at position D; HP = head plate; ILPI, ILPII = intersegmental lateral plates I–II; MPI, MPII, MPIII = median plates I–III (a: anterior, b: posterior); NP = neck plate; PPII, PPIII = segmental paired plates II–III; SP = pseudosegmental plate; ScLP = scapular segmental lateral plates; SeLPII, SeLPIII = segmental lateral plates II–III; SP = scapular plate (a: anterior, b: posterior); TP = terminal plate.

all dorsal plates covered with coarse, uniformly distributed granules which consist of hemispherical cuticular thickenings; posterior half of head plate and neck plate only with very fine, densely distributed punctations which consist of pillar structures of epicuticle; and small region around primary papilla and cirrophore of cirrus A devoid of any sculpture. Granules larger on segmental paired plates II and III, and pseudosegmental plate, ca. 1.5 μ m in diameter; granules medium in size on anterior half of head



Fig. 2. *Pseudechiniscus asper*, new species, holotype male (NSMT-Tg 44), Hokkaido, N Japan, differential interference contrast photomicrographs: A, Habitus, dorsal view; B, Internal claw of leg II; C, Detail of segmental paired plate III. Scales = 50 μ m (A), 10 μ m (B, C).

plate, scapular plate, median plates, and terminal plate, ca. 0.9 μ m in diameter; granules smaller on most anterior part of scapular plate, and intersegmental lateral plates I and II, ca. 0.5 μ m in diameter. Granules larger in central region of each plate than those in peripheral region of same plate. Space between granules smooth, without striae.

Head plate (HP) well-defined, gently faceted, subdivided into smaller pieces by smaller anterior and larger posterior Wshaped sculptures; ridge of large W-shaped sculpture effaced in its middle. Most posterior points of large W-shaped sculpture and minuscule indentations of neck plate interconnected by rather shallow, inconspicuous cuticular furrow which is dilated anteriorly. Neck plate (NP) narrow but appreciable; almost entire neck plate overlapping most posterior part of head plate; posterior edge of neck plate with a pair of vague, minuscule indentations.

Scapular plate (=plate I; SP) well developed, inconspicuously subdivided at middorsal line of body, shallowly subdivided into anterior and posterior subrectangular parts (SPa, SPb); anterior part fairly larger than posterior part. Three pairs of scapular segmental lateral plates (ScLP) separated from central main part; anterior segmental lateral plate connected with anterior central part; two pairs of posterior segmental lateral plates arranged transversely, connected with posterior central part.

Median plate I (MPI) distinctly subdivided into anterior and posterior parts (MPIa, MPIb); anterior part subrectangular, fairly larger, somewhat wider than posterior part, inconspicuously but completely subdivided at middorsal line of body; posterior part separated from anterior part as a transverse strip, concave dorsally, undivided at middorsal line of body; anterior and posterior edges of median plate I slightly and moderately angulated at middorsal line of body respectively.

Intersegmental lateral plate I (ILPI) subdivided into anterior and posterior parts; anterior part wider than posterior part; proximal edge of two parts obtuse.

Segmental paired plate II (PPII) well developed, with two pairs of lateral segmental plates (SeLPII); posterior segmental lateral plate somewhat larger than anterior segmental lateral plate; central main part of segmental paired plate II with cuticular ridges (cr) laterally; ridges effaced centrally forming separate anterior and posterior lines. Cuticular ridge, which is made up by posterior edge of segmental plate II and anterior edge of intersegmental lateral plate II, forming blunt swelling in its most distal part.

Median plate II (MPII) larger than median plates I and III, distinctly subdivided into anterior and posterior parts (MPIIa, MPIIb); anterior part fairly larger and wider than posterior part, weakly subdivided at middorsal line of body; posterior part forming a narrow transverse strip, concave dorsally, undivided at middorsal line of body; anterior and posterior edges of median plate II angulated at middorsal line of body.

Intersegmental lateral plate II (ILPI) subdivided into anterior and posterior parts; anterior part larger than posterior part; proximal points of two parts acute.

Segmental paired plate III (PPIII) well developed, similar to segmental paired plate II in size and shape, with two pairs of lateral segmental plates (SeLPIII); anterior and posterior lateral segmental plates incompletely separated; central main part of segmental paired plate III with cuticular ridges (cr) laterally; ridges effaced centrally forming separate anterior and posterior lines. Projection at position D (D) subtriangular, broad-based, ca. 9 μ m long.

Median plate III (MPIII) well-defined, rhomboid, undivided; anterior and posterior edges of median plate III moderately and poorly angulated at middorsal line of body, respectively.

Pseudosegmental plate (PsP) distinctly and wholly subdivided at middorsal line of body; paired lobes roundly triangular, each tipped with papillate projection, 8 μ m high, 17 μ m wide; lobes slightly conjoined middorsally with each other; posterior edge of pseudosegmental plate weakly arcuate but not sinuate. Posterior edge of pseudosegmental plate covering anterolateral side of terminal plate swollen in appearance.

Terminal plate (TP) with remarkable, long cuticular ridge posterolaterally, thus having a strongly facetted appearance, and with very blunt cuticular ridge on its middorsal line of body.

Subcephalic plate thin but comparably conspicuous, situated below mouth opening; anterior edge moderately marked but effaced in its middle, distinctly arcuate; lateral edge well marked, arcuate, dilated anteriorly; posterior edge scarcely marked, arcuate. Dense patches of cuticular granulation situated between each pair of legs I-III, and circum-genital region; each granule similar to that on dorsal plates, but much smaller; granules on central part of each patch somewhat larger than those on peripheral part; patch above gonopore with weakly developed edge, thus having a plate-like appearance, other patches without perceptible edge; patches between legs increasing in size posteriorly. Net pattern on ventral surface below subcephalic plate throughout but barely visible.

Leg plates developed on outer surfaces of legs I–III and dorsal surface of leg IV; each leg plate covered with granules similar to those on dorsal plates; granules larger but sparse on central part of leg plate, smaller but dense on peripheral part. Plate of leg IV without dentate collar. Sensory organ absent from legs I-III, not detected on leg IV.

Internal and external cirri and secondary clava situated ventrally. Internal and external cirri filamentous, without true cirrophore but with swollen base, without anchor-shaped bifurcation at tip; external cirrus longer than internal cirrus, 12 μ m long and 8 μ m long, respectively. Secondary clava hemispherical, situated nearer to external cirrus than to internal cirrus.

Primary clava and cirrus A situated anterolaterally on scapular plate; primary clava papillate, situated just beneath base of cirrus A, ca. 1.5 μ m long; cirrus A filamentous, short, with normal subconical cirrophore but without bifurcation at tip, ca. 20 μ m long, 12% of body length.

Claw normally curved distally, thickened basally; internal claw slightly longer than external claw on all legs; claws of leg I–III ca. 9 μ m long; claws of leg IV longer than claws of other legs, ca. 11 μ m. External claw on all legs without spur; each internal claw on all legs with a distinct, moderately curved spur directed toward base of claw; spur located at a height approximately ¼ as long as claw length; apex of spur not reaching base of claw.

Mouth opening situated ventrally. Buccal tube and stylet support weakly sclerotized, thus hardly discernible. Pharyngeal bulb ovoid, 14 μ m long, 11 μ m wide; outer cuticular lining inside pharyngeal bulb clearly thicker than bar-shaped placoid.

Male gonopore simple, being a slightly raised ring; anus normal, situated between leg IV of both sides.

Etymology.—From the Latin word *asper* (rough), referring to the general appearance of dorsal plates in the present new species.

Remarks.—In *Pseudechiniscus asper*, described in this paper, the single specimen collected is male. *Pseudechiniscus* males are very rare and have been reported only two times, viz. Dastych (1987) as *P. alberti* and Kristensen (1987) as unidentified species. Kristensen (1987) stated that *Pseudechiniscus* species with males are found

limnic or in other stable biotopes and that this is related to the thin sclerotization of the dorsal plates and the absence of the leg plates. It might be true in the species studied by Kristensen (1987), but it is remarkable that both *P. alberti* from Canada and *P. asper* from Japan were found in unstable environments such as terrestrial moss or lichen and have thick dorsal plates. Additional data are required to reveal the relationship between environmental habitat and the mode of reproduction.

Kristensen (1987) stated that all *Pseu*dechiniscus species are provided with the sensory organ on leg I. However, there are some species lacking this character, e.g., *P. juanitae* in Maucci (1986), *P. suillus* in Ramazzotti & Maucci (1983), Dastych (1989), and Kathman & Dastych (1990), *P. victor* in Maucci (1986), and *P. cfr. suillus* in McInnes (1995). Also, in *P. asper* the sensory organ on leg I is absent.

The sensory organ on leg IV, which is usually developed as a papilla or spine on its outer surface in *Pseudechiniscus* (Kristensen 1987), is not detected in the new species. This character is sometimes hard to observe, depending on the direction of the specimen on the glass slide, and we could not decide whether this character is absent or present in *P. asper*.

The large size of granules on dorsal plates, the presence of a subtriangular projection at the position D, and the presence of segmental lateral plates in the scapular plate and segmental paired plates II-III distinctly separate the new species from species such as Pseudechiniscus brevimontanus Kendall-Fite & Nelson, 1996, P. nataliae Biserov & Maucci in Biserov, 1986, P. ramazzottii Maucci, 1952, P. scorteccii Franceschi, 1952. P. asper bears some resemblance to P. occultus Dastych, 1980 from Poland, especially in the general appearance of the granulation on the dorsal plates, but can be readily distinguished from it mainly by the absence of the posterior lobe on the scapular plate, the absence of a spurs on the external claws of leg IV, and the absence of anchor-shaped bifurcations of cephalic cirri and cirrus A. The last character was omitted in the original description of *P. occultus* (Dastych 1980) but later emended as presented in Dastych (1987, 1988).

Acknowledgments

We would like to express our gratitude to Dr. Hieronymus Dastych of the Zoologisches Institut und Zoologisches Museum der Universität Hamburg for critically reading the first draft of this manuscript, and to Mr. Yoshihito Ohmura of the Graduate School of Science, The University of Tokyo for identifying the lichen sample. We are also grateful to anonymous reviewers who kindly made invaluable suggestions.

Literature Cited

- Biserov, V. I. 1986. Terrestrial water bears from the North Caucasus. 1. Heterotardigrada.—Zoologicheskii Zhurnal 65:747–756. [In Russian with English summary].
- Dastych, H. 1980. Niesporczaki (Tardigrada) Tatrzańskiego Parku Narodowego. [Tardigrades from the Tatra National Park].—Monografie Fauny Polski 9:1–232. [In Polish with English summary].

. 1987. Two new species of Tardigrada from the Canadian Subarctic with some notes on sexual dimorphism in the family Echiniscidae.— Entomologische Mitteilungen aus dem Zoologischen Staatsinstitut und Zoologischen Museum Hamburg 8:319–334.

- - . 1989. An annotated list of Tardigrada from the Antarctic.—Entomologische Mitteilungen

aus dem Zoologischen Museum Hamburg 9: 249–258.

- , & R. M. Kristensen 1995. Echiniscus ehrenbergi sp. n., a new water bear from the Himalayas (Tardigrada).—Entomologische Mitteilungen aus dem Zoologischen Museum Hamburg 11:221–230.
- Franceschi, T. 1952. Sul ritrovamento in Valcamonica di *Pseudechiniscus novaezeelandiae* f. marinae Bartos e di *Pseudechiniscus scorteccii* n. sp. (Tardigrada).—Doriana 1(30):1–7.
- Kathman, R. D., & H. Dastych. 1990. Some Echiniscidae (Tardigrada: Heterotardigrada) from Vancouver Island, British Columbia, Canada.—Canadian Journal of Zoology 68:699–706.
- Kendall-Fite, K., & D. R. Nelson 1996. Two new species of tardigrades from Short Mountain, Tennessee, USA.—Zoological Journal of the Linnean Society 116:205–214.
- Kristensen, R. M. 1987. Generic revision of the Echiniscidae (Heterotardigrada), with a discussion of the origin of the family. Pp. 261–335 in R. Bertolani, ed., Biology of Tardigrades: Proceedings of the 4th International Symposium on the Tardigrada, Modena, September 3–5, 1985.—Collana U.Z.I. Selected Symposia and Monographs 1, Mucchi Editore, Modena, 380 pp.
- Maucci, W. 1952. Un nuovo *Pseudechiniscus* del Carso Triestino (Tardigrada, Scutechiniscidae).— Atti della Società Italiana di Scienze Naturali e del Museo Civico di Storia Naturale in Milano 91:127–130.
- ———. 1986. Tardigrada.—Fauna d'Italia 24, Edizioni Calderini, Bologna, 388 pp.
- McInnes, S. J. 1995. Tardigrades from Signy Island, South Orkney Islands, with particular reference to freshwater species; Signy Island; South Orkney; Antarctica.—Journal of Natural History 29:1419–1445.
- Ramazzotti, G., & W. Maucci 1983. Il philum Tardigrada: III edizione riveduta e aggiornata.—Memorie dell'Istituto Italiano di Idrobiologia 41:1– 1012.